

NUTRITION, FEEDING, AND CALVES

The Prediction of Methane Production of Holstein Cows by Several Equations¹

V. A. WILKERSON and D. P. CASPER²

Energy Metabolism Unit
Nutrient Conservation and Metabolism Laboratory
Livestock and Poultry Sciences Institute
USDA, Agricultural Research Service
Beltsville, MD 20705

D. R. MERTENS³

US Dairy Forage Research Center
USDA, Agricultural Research Service
Madison, WI 53706

ABSTRACT

Ruminants are one of many sources contributing to atmospheric methane. The accuracy of seven published equations for methane prediction was evaluated using a data file consisting of 16 experiments (602 observations). Methane energy emissions ranged from .89 to 7.21 Mcal/d for Holstein cows. The DMI ranged from 9.7 to 28.7 kg/d for lactating cows and 4.0 to 12.9 kg/d for nonlactating cows. Mean dietary concentrations of ADF, CP, and ether extract were similar for lactating and nonlactating cows (20.9, 16.5, and 3.0% for lactating cows versus 21.2, 15.7, and 2.9% for nonlactating cows, respectively). Milk production ranged from 2.7 to 55.9 kg/d. Prediction equations were ranked by correlation coefficients and error of prediction. Prediction of methane energy loss from lactating and nonlactating Holstein cows with equations based on the daily total intake or intake of digested cellulose, hemicellulose, and nonfiber carbohydrates (OM - NDF - CP - ether

extract) provided the highest correlation coefficients for reproducibility and the lowest errors of prediction. Predictions were poor for lactating cows when a quadratic function of DMI was used. In general, equations estimated methane production more accurately and precisely for nonlactating than for lactating cows.

(Key words: methane, cattle, environment, energy loss)

Abbreviation key: EMU = Energy Metabolism Unit, NFC = nonfiber carbohydrate, *p* = concordance correlation coefficient (parenthetical use).

INTRODUCTION

The concentration of methane in the atmosphere has doubled over the past two centuries, which has been attributed to increasing emissions from human-related activities. When the number of wild buffalo and other ruminants in America before 1800 and the current decline in the dairy cow population are taken into consideration, it is difficult to conclude that ruminants are responsible for the increase in atmospheric methane. However, methane emissions from domesticated livestock in the US were estimated to range from 4.6 to 6.9×10^{12} g during 1990, and ruminants accounted for 95% of these emissions (24). Dairy cows, beef cows, and feedlot cattle were estimated to produce 4.4, 8.0, and 4.0% of total methane emissions in the US, respectively (24). These classes of livestock have been targeted by the US Environmental Protection Agency for reductions in methane

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²Present address: Cargill Animal Nutrition Center, PO Box 301, Elk River, MN 55330.

³Reprint requests.

emissions. The ability of the US Environmental Protection Agency to estimate the contributions that ruminants make to methane emissions and to recommend reduction in methane emissions depends on the accuracy of predicting methane production by ruminants.

Fermentation in anaerobic environments results in the production of hydrogen during the digestion of nutrients, especially carbohydrates. Methanogenic bacteria utilize this excess hydrogen to reduce carbon dioxide to methane. The symbiosis between bacteria that ferment carbohydrates and the methanogens, such as *Methanobrevibacter ruminantium* and *Methanomicrobium mobilis* (20), results in increased digestion and microbial production. The stoichiometry of VFA production during ruminal fermentation indicates that processes resulting in increased VFA and a shift in ratios toward acetate and butyrate production result in greater methane production by ruminants. In addition, feed ingredients that are high in fat, which is not fermented in the rumen, may also inhibit ruminal fermentation, resulting in decreased methane production by dairy cattle.

Methane production by ruminants has been predicted using equations involving DMI (2, 13); intake of carbohydrates (18); intake of digested carbohydrates (5, 18); digestibility and intake of dietary energy (4); and animal size, milk components, intake, and digestibility of dietary components (10). The variables used in these prediction equations correspond to biological relationships among animal, diet, and ruminal fermentation. Dry matter intake is a reasonable measure of the potentially fermentable substrate that is consumed by ruminants and then fermented to produce methane. Intake of carbohydrates or digested carbohydrates should be a more reliable measure of fermentable substrate than is DMI. Because these equations have been used to estimate methane production by domesticated livestock (24), they should be compared to determine their relative accuracy. The objective of this study was to evaluate the accuracy of published equations in predicting methane production using a data file compiled from indirect respiration experiments conducted at the Energy Metabolism Unit (EMU) of the Nutrient Conservation and Metabolism Laboratory at Beltsville, Maryland.

MATERIALS AND METHODS

Methane prediction equations vary from a simple equation using DMI to a complex system of equations based on physiological status and supplemental fat (Table 1). Published equations for predicting methane production include one each from Axelsson (2), Blaxter and Clapperton (4), Bratzler and Forbes (5), and Kriss (13) and two from Moe and Tyrrell (18). The approach used by Holter and Young (10) to predict methane energy involved the use of three equations, each used for a specific class of dairy cows: nonlactating cows, lactating cows not fed supplemental dietary fat, and lactating cows fed supplemental dietary fat. When using the system of Holter and Young (10), we assumed that any diet containing 3.5% or more of ether extract included supplemental fat. The equation of Blaxter and Clapperton (4) and the equation of Holter and Young for nonlactating cows (10) were incorrect as published, and the corrected equations are given in Table 1.

The entire EMU database contains balance trials conducted between 1960 and 1992. However, only 16 experiments conducted between 1976 and 1992 were included in the methane data file that was used to evaluate prediction equations because experiments between 1960 and 1976 were used by Moe and Tyrrell (18) to develop their equations. Balance trials retained in the methane data file were selected to contain all variables required by the prediction equations that were evaluated. This selection allowed all prediction equations to be evaluated using the same methane data file. The data file contained 602 energy balance trials utilizing 188 Holstein cows that ranged in age from 2 to 12 yr. Diets were typical TMR that ranged in forage concentration from 40 to 60%. Forage or fiber sources included alfalfa and orchardgrass as hays or silages, corn silage, beet pulp, and cottonseed hulls. Energy ingredients included barley, corn meal, corn earlage, oats, dried whey, and high moisture corn. Blood meal, dried brewers grains, corn gluten meal, cottonseed meal, distillers dried grains, fish meal, linseed meal, and soybean oil meal represented the protein ingredients. Supplemental dietary fat was provided by cocoa butter, cottonseed, Megalac® (Church and Dwight Co., Inc., Princeton, NJ), safflower oil, shortening, soybeans, and sunflower oil.

A general description of the EMU and the open circuit indirect respiration chambers has been reported by Flatt et al. (7). Composite samples of rations, Orts, and feces were dried at 65°C and ground through a 1-mm screen. The DM of rations, Orts, and feces was determined by drying at 105°C in a forced-air oven. Fiber analyses were based on the methods of Goering and Van Soest (8), except that sodium sulfite was omitted from NDF analysis. Non-fiber carbohydrates (NFC) were calculated using the formula: $NFC = OM - NDF - CP - \text{ether extract}$. This fraction was originally defined as soluble residue by Moe and Tyrrell (18), but NFC more clearly describes this dietary component (16, 17). Energy was determined by combustion of fresh rations, Orts, urine, and feces in an adiabatic oxygen bomb calorimeter with the aid of a polyethylene bag primer of known weight and energy content. Urine energy was determined from samples evaporated to near dryness at 40°C in a forced-

air oven. Milk energy was determined similarly without a primer after samples were evaporated to near dryness at 40°C in a forced-air oven. Nitrogen was determined by macro-Kjeldahl (1) and crude fat by ether extraction (1). Ether extract determination was by one of two methods, crucible weight gain or sample weight loss following ether reflux. The ether extraction method was always the same within experiments.

Formulas used to compute balances of energy, C, and N were reported by Flatt and Tabler (6). The EMU database contained most of the variables needed by the prediction equations. Additional variables needed by some of the equations were derived from variables within the methane data file. Digested carbohydrate (5) was calculated as the sum of digested NFC and digested NDF. Intake, as a multiple of maintenance energy needs (4), was calculated by dividing the intake of metabolizable energy by the metabolizable energy re-

TABLE 1. Equations used to predict methane production by Holstein cows.

Kriss (13)

$$\text{Methane (Mcal/d)} = (18 + 22.5 \times \text{DMI (kg/d)}) \times .013184 \text{ (Mcal/g of methane)}$$

Axelsson (2)

$$\text{Methane (Mcal/d)} = -.494 + .629 \times \text{DMI (kg/d)} - .025 \times \text{DMI}^2 \text{ (kg/d)}$$

Blaxter and Clapperton (4)

$$\begin{aligned} \text{Methane (Mcal/d)} = & (1.30 + .112 \times \text{energy digestibility determined at maintenance intake (\% of gross energy)} \\ & + \text{multiple of maintenance} \times (2.37 - .050 \times \text{energy digestibility at maintenance intake (\% of gross energy)})) \\ & + 100 \times \text{gross energy intake (Mcal/d)} \end{aligned}$$

Bratzler and Forbes (5)

$$\text{Methane (Mcal/d)} = (17.68 + .04012 \times \text{digested carbohydrate (g/d)}) \times .013184 \text{ (Mcal/g of methane)}$$

Moe and Tyrrell (18)

Intake of carbohydrate fractions

$$\text{Methane (Mcal/d)} = .814 + .122 \times \text{nonfiber carbohydrate (kg/d)} + .415 \times \text{hemicellulose (kg/d)} + .633 \times \text{cellulose (kg/d)}$$

Intake of digested carbohydrate fractions

$$\text{Methane (Mcal/d)} = .439 + .273 \times \text{digested nonfiber carbohydrate (kg/d)} + .512 \times \text{digested hemicellulose (kg/d)} + 1.393 \times \text{digested cellulose (kg/d)}$$

Holter and Young (10)

Nonlactating cows

$$\begin{aligned} \text{Methane (Mcal/d)} = & (12.12 - .00542 \times \text{BW (kg)} - .0900 \times \text{ADF (\% DMI)} + .1213 \times \text{ADF digestibility (\%)} \\ & - 2.472 \times \text{digestible energy (Mcal/kg of DM)} + .0417 \times \text{neutral detergent solubles digestibility (\%)} - .0748 \\ & \times \text{cellulose digestibility (\%)} + .0339 \times \text{hemicellulose digestibility (\%)} + 100 \times \text{gross energy intake (Mcal/d)}) \end{aligned}$$

Lactating cows fed supplemental dietary fat

$$\begin{aligned} \text{Methane (Mcal/d)} = & (2.898 - .0631 \times \text{milk (kg/d)} + .297 \times \text{milk fat (\%)} - 1.587 \times \text{milk protein (\%)} + .0891 \\ & \times \text{CP (\% DM)} + .1010 \times \text{forage ADF (\% DM)} + .102 \times \text{DMI (kg/d)} - .131 \times \text{ether extract (\% DM)} + .116 \\ & \times \text{DM digestibility (\%)} - .0737 \times \text{CP digestibility (\%)} + 100 \times \text{gross energy intake (Mcal/d)}) \end{aligned}$$

Lactating cows not fed supplemental dietary fat

$$\begin{aligned} \text{Methane (Mcal/d)} = & (2.927 - .0405 \times \text{milk (kg/d)} + .335 \times \text{milk fat (\%)} - 1.225 \times \text{milk protein (\%)} + .248 \\ & \times \text{CP (\% DM)} - .448 \times \text{ADF (\% DM)} + .502 \times \text{forage ADF (\% DM)} + .0352 \times \text{ADF digestibility (\%)} \\ & + 100 \times \text{gross energy intake (Mcal/d)}) \end{aligned}$$

quired for maintenance, which were estimated to be 119.7 kcal/kg of BW^{.75} (19). This maintenance energy requirement was derived by subtracting the 10% activity allowance from the NRC (19) value (133 kcal/kg of BW^{.75}) because the activity of cows in the calorimeters was limited. Forage ADF as a percentage of DMI was calculated for the equation of Holter and Young (10). The equations of Bratzler and Forbes (5) and Kriss (13) were converted from daily grams of methane to daily megacalories of methane by multiplying them by .013184 (.001 \times 9.45 kcal/L of methane divided by .7168 g/L of methane).

The equations of Blaxter and Clapperton (4) and Holter and Young (10) were converted from the percentage of gross energy lost as methane to daily megacalories of methane production by multiplying gross energy intake (kilograms of DMI per day times the megacalories of gross energy per kilogram of DM). The independent variables used in the prediction equations are listed, and mean, minimum, and maximum values for lactating and nonlactating cows are given in Table 2.

Scatter plots fitted with least squares regression lines (21) and a one-to-one line were used to indicate the relationship between predicted

TABLE 2. Mean, minimum, and maximum values of variables used to evaluate methane prediction equations for lactating and nonlactating Holstein cows.

| Variable | Lactating cows | | | Nonlactating cows | | |
|------------------------------|----------------|---------|---------|-------------------|---------|---------|
| | Mean | Minimum | Maximum | Mean | Minimum | Maximum |
| Cows, no. | 382 | ... | ... | 220 | ... | ... |
| BW, kg | 614 | 426 | 841 | 670 | 490 | 852 |
| Milk | | | | | | |
| kg/d | 28.0 | 2.7 | 55.9 | | | |
| Fat, % | 3.6 | 1.7 | 5.5 | | | |
| Protein, % | 3.2 | 2.6 | 4.4 | | | |
| Diet | | | | | | |
| CP, % | 16.5 | 10.3 | 22.1 | 15.7 | 10.5 | 20.7 |
| Ether extract, % | 3.0 | 1.6 | 6.5 | 2.9 | 1.6 | 5.7 |
| ADF, % | 20.9 | 14.9 | 34.5 | 21.2 | 14.7 | 36.3 |
| Gross energy, Mcal/kg | 4.60 | 4.32 | 5.96 | 4.55 | 4.11 | 4.90 |
| Digestible energy, Mcal/kg | 3.07 | 2.46 | 3.55 | 3.30 | 2.69 | 3.79 |
| Intake | | | | | | |
| DM, kg/d | 18.72 | 9.68 | 28.66 | 6.53 | 3.98 | 12.89 |
| Multiples of maintenance | 3.39 | 1.55 | 5.20 | 1.11 | .62 | 2.37 |
| Nonfiber carbohydrates, kg/d | 7.09 | 2.53 | 11.07 | 2.48 | .63 | 5.52 |
| Hemicellulose, kg/d | 2.80 | 1.02 | 7.62 | 1.01 | .38 | 2.72 |
| Cellulose, kg/d | 2.97 | 1.13 | 5.82 | 1.06 | .56 | 3.14 |
| Gross energy, Mcal/d | 86.3 | 43.2 | 132.8 | 29.7 | 18.2 | 59.9 |
| Digested, kg/d | | | | | | |
| Nonfiber carbohydrates | 6.39 | 2.05 | 10.30 | 2.33 | .58 | 4.86 |
| NDF | 3.13 | .83 | 8.76 | 1.30 | .43 | 3.53 |
| Hemicellulose | 1.40 | .22 | 5.80 | .62 | .15 | 1.77 |
| Cellulose | 1.52 | .54 | 4.14 | .64 | .27 | 1.94 |
| Digestibility, % | | | | | | |
| DM | 66.9 | 56.8 | 74.8 | 72.2 | 60.5 | 79.8 |
| CP | 65.1 | 42.7 | 75.5 | 70.4 | 52.0 | 82.0 |
| Neutral detergent solubles | 81.6 | 65.8 | 91.3 | 86.7 | 76.4 | 98.9 |
| NDF | 45.3 | 19.7 | 67.0 | 53.8 | 27.2 | 77.5 |
| ADF | 43.3 | 20.5 | 67.0 | 48.6 | 19.1 | 68.2 |
| Hemicellulose | 47.3 | 16.3 | 76.2 | 60.8 | 28.8 | 86.3 |
| Cellulose | 50.4 | 25.5 | 71.7 | 60.6 | 37.7 | 77.9 |
| Forage, kg/d | | | | | | |
| ADF | 4.51 | 1.30 | 10.49 | 1.29 | .67 | 3.51 |
| Orts, kg/d | | | | | | |
| Cows, no. | 308 | ... | ... | 7 | ... | ... |
| ADF | .79 | .002 | 3.21 | .16 | .02 | .39 |

and observed production of methane. Mean absolute error of prediction and regression standard error of prediction were obtained to indicate the magnitude of prediction error from the observed and regression lines, respectively. The mean absolute error of prediction was calculated as the sum of the absolute value between predicted and observed methane production divided by the number of observations. The regression standard error of prediction was calculated as the square root of the mean squared error. The Pearson correlation coefficient (21) and the concordance correlation coefficient (p) (14, 15) were calculated for each equation. The Pearson correlation coefficient indicates the precision of prediction in relation to the regression equation between predicted and observed methane production. However, an equation with high precision can have low accuracy in predicting methane production because of prediction bias, which can be either constant or systematic. A bias correction factor and the Pearson correlation coefficient are used in the concordance correlation coefficient to determine the total accuracy in predicting methane production. The concordance correlation coefficient indicates the accuracy of prediction in relation to the one-to-one line between predicted and observed methane production. Methane prediction equa-

tions were ranked according to their repeatability in estimating the observed methane production with the concordance correlation coefficient (14, 15).

RESULTS AND DISCUSSION

The EMU methane data file consists of 382 observations for lactating cows and 220 observations for nonlactating cows (Table 2). Lactating cows had lower mean BW with a wider range than nonlactating cows. As expected, DMI and intake of dietary components were greater for lactating cows than nonlactating cows. In 14 of the 16 experiments, nonlactating cows were fed the ration offered to lactating cows at an intake near maintenance. Thus, rations for nonlactating cows in the EMU database differed from typical rations for nonlactating cows because they were higher in concentrates.

Observed methane production averaged 3.81 Mcal/d for all cows in the methane data file (Table 3, Observed), which agreed with the projected mean of 3.88 Mcal/d of methane emission from herds of lactating and nonlactating dairy cows in Japan (22), although our data file contained a greater proportion of nonlactating cows than would be expected in typical dairy herds. Methane production (Table 3) by

TABLE 3. Methane production of lactating, nonlactating, and all Holstein cows from an observed data file and predicted methane production using seven literature sources.

| Equation ¹ | Lactating cows (n = 382) | | | Nonlactating cows (n = 220) | | | All cows (n = 602) | | |
|-----------------------|-----------------------------|---------|---------|--------------------------------|---------|---------|-----------------------|---------|---------|
| | Mean | Minimum | Maximum | Mean | Minimum | Maximum | Mean | Minimum | Maximum |
| | (Mcal/d) | | | | | | | | |
| Observed | 4.70 | 1.46 | 7.21 | 2.28 | .89 | 4.29 | 3.81 | .89 | 7.21 |
| [1] | 5.79 | 3.11 | 8.74 | 2.17 | 1.42 | 4.06 | 4.47 | 1.42 | 8.74 |
| [2] | 2.21 | -3.00 | 3.46 | 2.42 | 1.61 | 3.46 | 2.29 | -3.00 | 3.46 |
| [3] | 4.65 | 3.19 | 6.50 | 2.34 | 1.52 | 4.07 | 3.81 | 1.52 | 6.50 |
| [4] | 5.27 | 3.04 | 7.61 | 2.15 | 1.35 | 3.79 | 4.13 | 1.35 | 7.61 |
| [5] | 4.72 | 2.51 | 7.03 | 2.21 | 1.55 | 4.25 | 3.80 | 1.55 | 7.03 |
| [6] | 5.01 | 2.71 | 8.39 | 2.29 | 1.40 | 4.60 | 4.02 | 1.40 | 8.39 |
| [7] | 4.77 | 1.61 | 8.04 | 1.61 | .38 | 3.42 | 3.61 | .38 | 8.04 |
| | (% of gross energy intake) | | | | | | | | |
| Observed | 5.49 | 2.53 | 7.82 | 7.89 | 3.47 | 10.07 | 6.37 | 2.53 | 10.07 |

¹Observed Energy Metabolism Unit methane data file; Equation [1], Kriss (13); Equation [2], Axelsson (2); Equation [3], Blaxter and Clapperton (4); Equation [4], Bratzler and Forbes (5); Equation [5], Moe and Tyrrell (18) using intake of carbohydrate fractions; Equation [6], Moe and Tyrrell (18) using intake of digest carbohydrate fractions; and Equation [7], Holter and Young (10).

lactating cows (4.70 Mcal/d) was higher than for nonlactating cows (2.28 Mcal/d), which was probably related to the lower intake of fermentable substrate by nonlactating cows than by lactating cows (Table 2). However, per unit of DMI, lactating cows produced less methane (.25 Mcal/kg of DM) than did nonlactating cows (.35 Mcal/kg of DM). The similar composition of diets for lactating and nonlactating cows suggests that something other than chemical composition of the ration influenced methane emissions. Slower rates of passage in nonlactating cows with low DMI should have resulted in more extensive ruminal fermentation and greater production of methane. The works of Bath and Rook (3) and Sutton et al. (23) suggest that slower rates of passage alter ruminal VFA patterns, which could increase methane production by ruminants when more acetate and butyrate are produced. Our values for methane production by lactating cows were 16, 36, and 10% greater than those reported by Kirchgessner et al. (12), Holter and Young (10), and Shibata (22), respectively, and methane production by nonlactating cows was 2 and 6% less than those reported by Holter and Young (10) and Shibata (22), respectively.

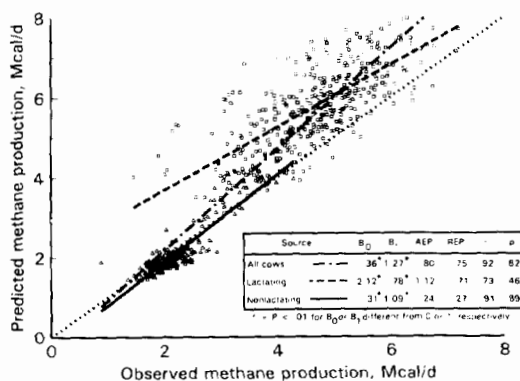


Figure 1. Predicted and observed methane production (megacalories per day) for lactating (\square) and nonlactating (Δ) Holstein cows using the equation of Kriss (13); \dots = one-to-one line for observed methane production, $---$ = least squares regression line for lactating cows, $-$ = least squares regression line for nonlactating cows, and $- \cdot -$ = least squares regression line for all cows. Statistics shown in tabular insert include B_0 = intercept, B_1 = slope of the regression of predicted versus observed methane production, AEP = absolute error of prediction, REP = regression standard error of prediction, r = Pearson correlation coefficient, and p = concordance correlation coefficient.

The equation by Kriss (13), which is based on DMI, was the simplest of the published equations for predicting methane production that were evaluated. This equation was derived from 131 observations using 4 cows and 20 steers (54 observations from roughage and 77 observations from roughage plus grain diets). Compared with data of Kriss (13), the EMU data file was five times larger and used eight times as many animals; however, a complete roughage diet was not part of the EMU data file.

The mean bias for each equation can be calculated by subtracting the mean methane production from mean predicted methane production for each equation (Table 3). The equation of Kriss (13) overestimated mean methane production (.66 Mcal/d) for all cows, because of a large overestimation for lactating cows (1.09 Mcal/d). The mean bias of this equation for nonlactating cows was small (.11 Mcal/d). The relationship between predicted and observed methane production for all cows (Figure 1) showed a systematic bias for the Kriss (13) equation, which is a good predictor when methane production is low, but increasingly overestimates when methane production is high. The high correlation coefficients ($r = .92$; $p = .82$) between predicted and observed methane production suggest that the regression equation between the two could be used to adjust for much of the systematic error in the Kriss (13) equation. The high regression error of prediction (19.7% of the mean; Figure 1) for all cows suggests that variables other than DMI are needed to improve the prediction accuracy.

The large mean absolute error of prediction (21.0% of the mean; Figure 1) indicated that the equation of Kriss (13) was not acceptable for direct prediction of methane production for all cows. Results shown in Figure 1 for nonlactating cows indicate that the Kriss (13) equation has a small systematic bias, which limited the ability of this equation to predict methane production by lactating cows (Figure 1).

The equation of Axelsson (2) is also based on DMI but uses linear and quadratic terms of DMI for predicting methane production. A total of 176 observations of adult cattle were used to develop this equation. Mean DMI was lower for the Axelsson (2) data file than for the EMU data file (8.58 vs. 14.26 kg/d, respectively).

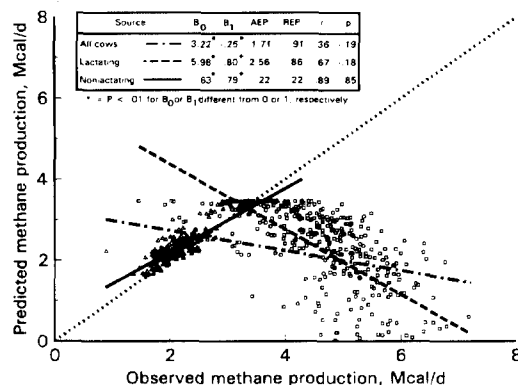


Figure 2. Predicted and observed methane production (megacalories per day) for lactating (\square) and nonlactating (\triangle) Holstein cows using the equation of Axelsson (2); ... = one-to-one line for observed methane production, ---- = least squares regression line for lactating cows, — = least squares regression line for nonlactating cows, and -.- = least squares regression line for all cows. Statistics shown in tabular insert include B_0 = intercept, B_1 = slope of the regression of predicted versus observed methane production, AEP = absolute error of prediction, REP = regression standard error of prediction, r = Pearson correlation coefficient, and p = concordance correlation coefficient.

The Axelsson (2) equation underestimated mean methane production (-1.52 Mcal/d) for all cows, which was caused by a large underestimation for lactating cows (-2.49 Mcal/d). The mean bias of this equation was small (.14 Mcal/d) for nonlactating cows. The systematic bias for the Axelsson (2) equation (Figure 2) resulted in overestimation at low methane productions and increasing underestimation as methane production increased from about 3.0 Mcal/d. As DMI increased beyond maximum methane production (12.58 kg of DMI/d, calculated from the first derivative of the quadratic equation), predictions of methane production were increasingly underestimated. The large mean absolute error of prediction (44.9% of the mean) indicated that the equation of Axelsson (2) was not acceptable for direct prediction of methane production for all cows because many observations would be outside the DMI of the data file used to derive the equation. The Axelsson (2) equation has a slight systematic bias for nonlactating cows (Figure 2) that overestimates at low methane production and underestimates at high methane production. Al-

ternatively, this same equation has a large systematic bias for lactating cows (Figure 2).

The equations of Kriss (13) and Axelsson (2) use constant coefficients to relate DMI to methane production. Blaxter and Clapperton (4) developed an equation that varied the relationship between DMI and methane production based on energy digestibility of the diet at maintenance and intake expressed as a multiple of maintenance. They defined maintenance as the metabolizable energy consumed at zero energy retention. Blaxter and Clapperton (4) utilized 391 observations involving 48 diets fed to sheep to determine the effects of amount and type of diet on methane production. The diets were classified as roughages, mixed diets, or pelleted diets, and regression analyses were performed on each class and on all diets. Blaxter and Clapperton (4) found similar regression coefficients among the classes of diets and recommended that the overall equation be used to predict methane production by ruminants.

Energy digestibility was measured at production intakes in the EMU data file. Assuming that digestibility at maintenance would be higher, our substitution of digestibility measured at production intakes for digestibility measured at maintenance in the Blaxter and Clapperton (4) equation resulted in a bias. At low intakes (low methane productions), the energy digestibilities measured in the EMU data file were at or near maintenance and comparable with that used by Blaxter and Clapperton (4). At high intakes (high methane productions), the energy digestibilities in the EMU data file would be lower than the energy digestibilities measured at maintenance by Blaxter and Clapperton (4) and result in higher estimates of methane production. However, using energy digestibilities measured at production intakes actually improved the fit of the Blaxter and Clapperton (4) equation.

The equation of Blaxter and Clapperton (4) has a mean bias of zero for all cows, which is a result of a small underestimation ($-.05$ Mcal/d) for lactating cows and an equally small overestimation (.06 Mcal/d) for nonlactating cows. The plot of predicted versus observed methane production for the Blaxter and Clapperton (4) equation (Figure 3) demonstrated a systematic bias for all cows with overestimations at low methane productions, compensating for underestimations at high methane

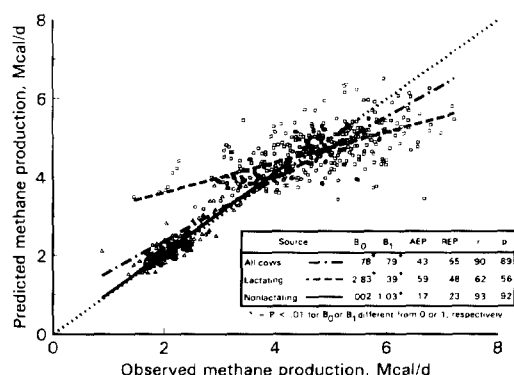


Figure 3. Predicted and observed methane production (megacalories per day) for lactating (\square) and nonlactating (\triangle) Holstein cows using the equation of Blaxter and Clapperton (4); \cdots = one-to-one line for observed methane production, $----$ = least squares regression line for lactating cows, $—$ = least squares regression line for the nonlactating cows, and $- - -$ = least squares regression line for all cows. Statistics shown in tabular insert include B_0 = intercept, B_1 = slope of the regression of predicted versus observed methane production, AEP = absolute error of prediction, REP = regression standard error of prediction, r = Pearson correlation coefficient, and p = concordance correlation coefficient.

productions. The high correlation coefficients ($r = .90$; $p = .89$) associated with all cow data suggest that a majority of the systematic prediction error can be removed by correcting predictions using a regression equation. The regression error of prediction for the Blaxter and Clapperton (4) equation was 14.4% of the mean methane production. The regression error was less than that of the equations by Kriss (13) and Axelsson (2), implying that prediction of methane production using energy digestibility of the diet and intake as a multiple of maintenance is an improvement over the equations using only DMI.

The absolute error of prediction for the Blaxter and Clapperton (4) equation was 11.3% of the mean, indicating an improvement in predicting methane production over equations using only DMI. Data for nonlactating cows resulted in a systematic bias (Figure 3) that predicted well at low methane production, but slightly overestimated when methane production was high. The data for lactating cows resulted in a large systematic bias; low methane production was overestimated, and

high methane production was underestimated (Figure 3). The low correlation coefficients for lactating cows ($r = .62$; $p = .56$) suggest that more information is needed to explain methane production than energy digestibility of diet and intake as a multiple of maintenance.

Although methane production can result from fermentation of protein in anaerobic environments, the majority of methane comes from the fermentation of carbohydrates (9, 11). Lipids are not extensively fermented in the rumen; therefore, they do not contribute to methane production in ruminants. In addition, lipids may reduce the digestion of carbohydrates in the rumen (25), and unsaturated fatty acids serve as a hydrogen sink, both of which would reduce methane production. Thus, carbohydrates apparently are the predominant precursor for methane production in the rumen.

The stoichiometric relationship presented by Hungate (11) indicates that methane production is related to the amount and ratios of VFA produced in the rumen. Factors that contribute to higher ratios and amounts of acetate and butyrate result in increased methane production; high ratios of propionic acid result in decreased methane production. Factors that increase acetate production or proportion, such as increased fiber in the ration (3) or slower fractional rates of passage, should result in increased methane production. Conversely, factors that increase the proportions of propionate during fermentation, such as high dietary nonstructural carbohydrate (starches and sugars) concentrations, should decrease methane production by ruminants. Based on these biological relationships, methane production should be predicted more accurately if intakes of specific dietary components are used as predictors. Accuracy may also be improved if the amounts of digested dietary components, which presumably are proportional to those fermented, are used to predict methane production.

Bratzler and Forbes (5) related methane production to the amount of digested total carbohydrate. Their equation was developed from 130 observations utilizing steers and cows of beef and dairy breeds. The equation of Bratzler and Forbes (5) was developed using a narrower range of digested total carbohydrates (.9 to 5.8 kg/d) than that in the EMU data file (.9 to 8.4 kg/d for nonlactating and 2.8 to 19.1 kg/d for lactating Holstein cows).

The mean bias for the Bratzler and Forbes (5) equation was .32 Mcal/d; most of this bias was due to the larger mean bias for lactating cows (.57 Mcal/d) than for nonlactating cows (-.13 Mcal/d). The lower mean bias for nonlactating cows supports the use of this equation when intake of total digested carbohydrate approximates the range used to develop the equation. The plot of observed versus predicted methane production for (Figure 4) for lactating cows showed a systematic bias with large overestimations at low methane productions.

High correlation coefficients ($r = .92$; $p = .89$) were obtained by the Bratzler and Forbes (5) equation using the EMU data file, suggesting that the regression equation between predicted and observed methane production can be used to adjust for much of the systematic error for all cows. The regression error of prediction (17.3% of the mean) demonstrated improved prediction of methane production over equations based solely on DMI (2, 13). The regression error of prediction for the Bratzler and Forbes (5) equation was greater than that observed by the Blaxter and Clapperton (4) equation (17.3 vs. 14.4% of the mean), because of the greater error in predicting methane production of lactating cows than of nonlactating cows.

The moderate absolute error of prediction (13.6% of the mean; Figure 4) indicated no improvement over the Blaxter and Clapperton (4) equation for predicting methane production of all cows. Predictions for nonlactating cows (Figure 4) signified a small and relatively constant bias. However, the ability of the equation to predict methane production by lactating cows was limited because of a systematic bias.

Fermentation biochemistry suggests that carbohydrates may differ in the amount of methane produced in the rumen. Fibrous carbohydrates such as hemicellulose and cellulose typically result in higher proportions of acetate during fermentation than do NFC and should result in greater methane production. Moe and Tyrrell (18) used the intake of NFC, hemicellulose, and cellulose to predict methane production. Their equations were developed from a data file containing 404 observations from Holstein cows used in studies to investigate the effects of dietary protein, proportions of concentrate, different feed ingredients, and physical form of corn on net energy. Our EMU data

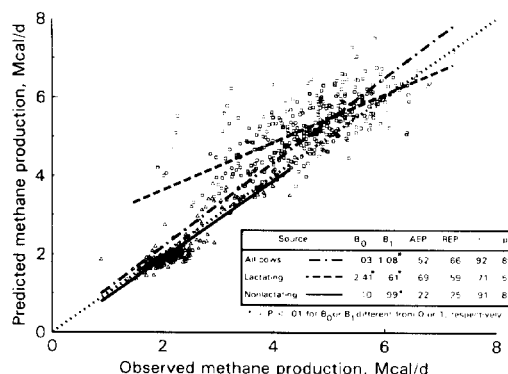


Figure 4. Predicted and observed methane production (megacalories per day) for lactating (\square) and nonlactating (Δ) Holstein cows using the equation of Bratzler and Forbes (5); ... = one-to-one line for observed methane production, --- = least squares regression line for lactating cows, — = least squares regression line for nonlactating cows, and - - - = least squares regression line for all cows. Statistics shown in tabular insert include B₀ = intercept, B₁ = slope of the regression of predicted versus observed methane production, AEP = absolute error of prediction, REP = regression standard error of prediction, r = Pearson correlation coefficient, and p = concordance correlation coefficient.

file (Table 2) had a wider range for carbohydrate fractions than did that of Moe and Tyrrell (18) (.95 to 9.66 kg/d of NFC, .47 to 4.41 kg/d of hemicellulose, and .41 to 3.90 kg/d of cellulose).

The mean bias for the equation of Moe and Tyrrell (18) using intake of carbohydrate fractions was a small underestimation at -.01 Mcal/d for all cows, resulting from the slight overestimation (.02 Mcal/d) for lactating cows and the slight underestimation (-.07 Mcal/d) for nonlactating cows. The predicted versus observed methane production relationship for all cows (Figure 5) showed a systematic bias for the Moe and Tyrrell (18) equation, which predicts well for the middle range of methane production, slightly overestimates at low methane production, and underestimates at high methane production. High correlation coefficients ($r = .92$; $p = .92$) for this equation suggest that the regression equation can be used to adjust for most of the systematic error in the equation of Moe and Tyrrell (18) using intake of carbohydrate fractions. The regression error of prediction for all cows was 15.0%

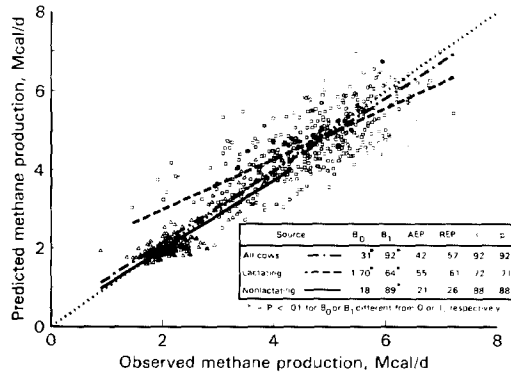


Figure 5. Predicted and observed methane production (megacalories per day) for lactating (\square) and nonlactating (Δ) Holstein cows using the intake of carbohydrate fractions equation of Moe and Tyrrell (18); \cdots = one-to-one line for observed methane production, $----$ = least squares regression line for lactating cows, $—$ = least squares regression line for nonlactating cows, and $---$ = least squares regression line for all cows. Statistics shown in tabular insert include B₀ = intercept, B₁ = slope of the regression of predicted versus observed methane production, AEP = absolute error of prediction, REP = regression standard error of prediction, r = Pearson correlation coefficient, and p = concordance correlation coefficient.

of the mean and was the lowest of the equations evaluated, suggesting that intake of carbohydrate fractions is valuable for accurate prediction of methane production by Holstein cows.

The mean absolute error of prediction (11.0% of the mean) for the equation by Moe and Tyrrell (18) using intake of carbohydrate fractions was the lowest of the equations evaluated across all cows. Results shown in Figure 5 for nonlactating cows indicate that this equation has a systematic bias that estimates well at low methane production but slightly underestimates at high methane production. This equation predicts methane production for lactating cows with less systematic bias than the equations of Kriss (13), Axelsson (2), and Blaxter and Clapperton (4).

The proportion of carbohydrate that is fermented in the gastrointestinal tract is a function of the fractional rate of passage, type, and source of carbohydrate. Therefore, Moe and Tyrrell (18) used digested NFC, digested hemicellulose, and digested cellulose to improve the predictions of methane production. This equation (18) resulted in a mean overesti-

mation of methane production (.21 Mcal/d) for all cows that was caused by a larger overestimation for lactating cows (.34 Mcal/d) than the small overestimation for nonlactating cows (.01 Mcal/d). The relationship of predicted versus observed methane production for the equation of Moe and Tyrrell (18) using intake of digested carbohydrate fractions (Figure 6) showed a slight systematic bias across the range of methane production by all cows in this study. The high correlation coefficients ($r = .93$; $p = .91$) of predicted versus observed methane production suggested that the regression equation between these two values can be used to adjust for the error in the equation of Moe and Tyrrell (18) using intake of digested carbohydrate fractions. The regression error of prediction (15.7% of the mean) supported the fractionation of total carbohydrate into digested NFC, digested hemicellulose, and digested cellulose to improve the prediction of methane production by Holstein cows.

The absolute error of prediction (12.1% of the mean) indicated that the equation of Moe and Tyrrell (18) using intake of digested carbo-

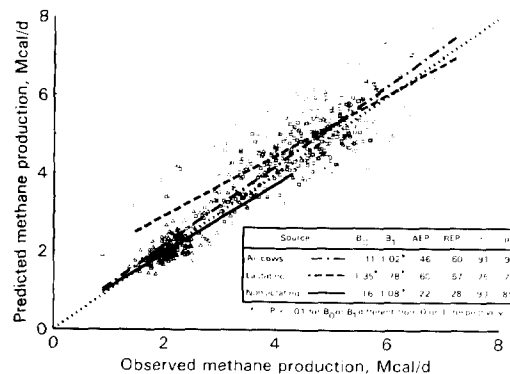


Figure 6. Predicted and observed methane production (megacalories per day) for lactating (\square) and nonlactating (Δ) Holstein cows using the intake of digested carbohydrate fractions equation of Moe and Tyrrell (18); \cdots = one-to-one line for observed methane production, $----$ = least squares regression line for lactating cows, $—$ = least squares regression line for nonlactating cows, and $---$ = least squares regression line for all cows. Statistics shown in tabular insert include B₀ = intercept, B₁ = slope of the regression of predicted versus observed methane production, AEP = absolute error of prediction, REP = regression standard error of prediction, r = Pearson correlation coefficient, and p = concordance correlation coefficient.

hydrate fractions is as accurate as the equation by Blaxter and Clapperton (4). The results shown in Figure 6 indicate that this equation has a small systematic bias for nonlactating cows.

Holter and Young (10) used a more empirical statistical approach to identify cow and dietary factors that influence methane production. They divided their data file into subsets to reduce cow and dietary effects. The subsets consisted of Holstein cows that included 60 observations for nonlactating cows, 107 observations for lactating cows fed supplemental dietary fat, and 90 observations for lactating cows not fed supplemental dietary fat.

Holter and Young (10) reported that methane production of nonlactating cows was positively related by the digestibility of ADF, neutral detergent solubles, cellulose, hemicellulose, and forage ADF in the ration. They also observed a negative relationship between methane production and dietary concentration of digestible energy. The equations of Holter and Young (10) use variables that are not precursors of methane production, such as milk production, milk fat, and milk protein (Table 1). Additionally, the equations of Holter and Young (10) have coefficients that are inconsistent with accepted relationships between diet and methane production. Equations for nonlactating cows and lactating cows not fed supplemental dietary fat contain negative coefficients for dietary ADF and positive coefficients for ADF digestibility, suggesting that rations with high ADF content would have less methane production than those with low ADF content. Similarly, the equation for lactating cows fed supplemental dietary fat has a positive regression coefficient for dietary CP and a negative regression coefficient for CP digestibility.

The mean prediction bias for all cows was an underestimation of .2 Mcal/d, which resulted from a slight overestimation (.07 Mcal/d) for lactating cows and a large underestimation (.67 Mcal/d) for nonlactating cows. The predicted versus observed relationship for this equation (Figure 7) indicates a systematic bias for all cows, which underestimates at low methane production and overestimates at high methane production. The absolute error of prediction was large (17.6%) for all cows using the combined predictions from the three equa-

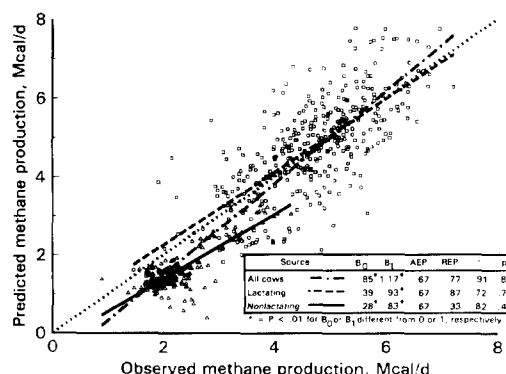


Figure 7. Predicted and observed methane production (megacalories per day) for lactating (\square) and nonlactating (\triangle) Holstein cows using the system of equations by Holter and Young (10); ... = one-to-one line for observed methane production, ---- = least squares regression line for lactating cows, — = least squares regression line for nonlactating cows, and - - - = least squares regression line for all cows. Statistics shown in tabular insert include B₀ = intercept, B₁ = slope of the regression of predicted versus observed methane production, AEP = absolute error of prediction, REP = regression standard error of prediction, r = Pearson correlation coefficient, and p = concordance correlation coefficient.

tions of Holter and Young (10). The correlation coefficients ($r = .91$; $p = .88$) suggest that prediction bias can be reduced using a regression of predicted versus observed methane production. The high regression error of prediction (20.2% of the mean; Figure 6) suggests that the equations of Holter and Young (10) are not as accurate as less complex equations and implies that their equations for prediction of methane production might be overparameterized.

Holter and Young (10) reported that increased percentages of fat in the ration reduced methane production by lactating cows and generated a separate prediction equation for this situation. We partitioned the EMU methane data file into lactating cows fed diets containing ether extract $\geq 3.5\%$ of DM ($n = 74$) and those receiving rations containing ether extract $< 3.5\%$ of DM ($n = 308$). The mean prediction bias was an overestimation (.08 Mcal/d) for lactating cows not provided supplemental dietary fat, and this bias was greater than the overestimation (.05 Mcal/d) for lactating cows receiving supplemental dietary fat.

Regressions between predicted and observed methane production demonstrated greater bias for lactating cows fed supplemental dietary fat (intercept: 1.30 ± 0 , $P < .02$; regression coefficient: $.75 \pm 1$, $P < .01$; data not shown) than lactating cows not fed supplemental dietary fat (intercept: $.28 \pm 0$, $P > .01$; regression coefficient: $.96 \pm 1$, $P < .01$; data not shown).

CONCLUSIONS

Many of the equations used to estimate methane production were evaluated outside of the range of data used to derive them. Although these equations may be more accurate when used within the range of the original data file, their use in predicting methane production may not be so restricted. Our review of published equations evaluated the possible errors and potential applicability of these equations for general use in estimating methane emissions by dairy cows.

The equation of Blaxter and Clapperton (4) for predicting methane production of nonlactating cows had the highest concordance correlation coefficient, which accounts for both accuracy and precision in methane emissions. Equations by Moe and Tyrrell (18), Bratzler and Forbes (5), Blaxter and Clapperton (4), Kriss (13), and Axelsson (2) were all adequate for predicting methane production by nonlactating Holstein cows. Errors of prediction associated with the data for nonlactating cows were much less than errors of prediction for lactating cows. In the EMU data file, nonlactating cows were fed rations offered to lactating cows at intakes restricted to near maintenance. There was a concern that feeding nonlactating cows under this regimen might bias methane production; however, the lack of bias across all equations indicated that this was not a problem.

Predictions of methane production were more variable for lactating cows than for nonlactating cows. The equation of Moe and Tyrrell (18) using intake of carbohydrate fractions ranked the highest for predicting methane production by lactating cows, based on its correlation coefficients and errors of prediction. The equation of Moe and Tyrrell (18) using intake of digested carbohydrate fractions and the equations of Holter and Young (10) had concordance correlation coefficients simi-

lar to the equation of Moe and Tyrrell (18) using intake of carbohydrate fractions for lactating cows, but both had greater errors of prediction.

The equation of Moe and Tyrrell (18) using intake of carbohydrate fractions was the most accurate and precise of the seven equations evaluated for the prediction of methane production by all cows. The carbohydrate variables, cellulose, hemicellulose, and NFC, used by Moe and Tyrrell (18) are relatively easy to obtain, which makes this equation usable by nutritionists and environmentalists for accurately predicting and monitoring reductions in ruminant methane emissions by the EPA. The equation of Moe and Tyrrell (18) using intake of digested carbohydrate fractions, the equations of Holter and Young (10) using a complex set of variables, the equation of Bratzler and Forbes (5) using intake of digested total carbohydrates, and the equation of Blaxter and Clapperton (4) using energy digestibility and intake as a multiple of maintenance could be used for the combined group of lactating and nonlactating Holstein cows with some loss of accuracy and precision. Further research in the area of methane production by Holstein cattle is needed to reduce prediction errors, especially for lactating cows.

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